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Search Results -

Term	Documents
CASSETTE.USPT.	55238
CASSETTES.USPT.	21010
(5 AND CASSETTE).USPT.	39
(L5 AND CASSETTE).USPT.	39

Database: US Pre-Grant Publication Full-Text Database; JPO Abstracts Database; EPO Abstracts Database; Derwent World Patents Index; IBM Technical Disclosure Bulletins

Search: L6

Refine Search

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Search History

DATE: Saturday, March 29, 2003 Printable Copy Create Case

Set Name side by side	Query	Hit Count	Set Name result set
	DB=USPT; PLUR=YES; OP=ADJ		
L6	L5 and cassette	39	L6
L5	l1 and (express$ same (mammalian or eukaryot$ or animal) same polymerase)	155	L5
L4	L1 and express$3 polymerase	12	L4
L3	L1 and express polymerase	0	L3
L2	L1 and "express polymerase"	0	L2
L1	@PRAY <= 1995 AND (polymerase same vector same express$)	811	L1

END OF SEARCH HISTORY

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Search Results - Record(s) 1 through 10 of 39 returned.

1. Document ID: US 6495666 B2

L6: Entry 1 of 39 File: USPT Dec 17, 2002

US-PAT-NO: 6495666
DOCUMENT-IDENTIFIER: US 6495666 B2

TITLE: Polypeptide composing human chimeric antibody

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | Claims | KWMC | Draw Desc | Image

2. Document ID: US 6440422 B1

L6: Entry 2 of 39 File: USPT Aug 27, 2002

US-PAT-NO: 6440422
DOCUMENT-IDENTIFIER: US 6440422 B1

TITLE: Recombinant MVA virus, and the use thereof

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWMC | Draw Desc | Image

3. Document ID: US 6437098 B1

L6: Entry 3 of 39 File: USPT Aug 20, 2002

US-PAT-NO: 6437098
DOCUMENT-IDENTIFIER: US 6437098 B1

TITLE: Human chimeric antibody specific for the ganglioside GD3

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWMC | Draw Desc | Image

4. Document ID: US 6426187 B1

L6: Entry 4 of 39 File: USPT Jul 30, 2002

US-PAT-NO: 6426187
DOCUMENT-IDENTIFIER: US 6426187 B1

TITLE: Detection of conversion to mucoidy in Pseudomonas aeruginosa infecting cystic

fibrosis patients

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc | Image

5. Document ID: US 6423511 B1

L6: Entry 5 of 39 File: USPT Jul 23, 2002

US-PAT-NO: 6423511
DOCUMENT-IDENTIFIER: US 6423511 B1

TITLE: Humanized antibodies

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc | Image

6. Document ID: US 6346414 B1

L6: Entry 6 of 39 File: USPT Feb 12, 2002

US-PAT-NO: 6346414
DOCUMENT-IDENTIFIER: US 6346414 B1

TITLE: Transposition assembly for gene transfer in eukaryotes

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc | Image

7. Document ID: US 6306652 B1

L6: Entry 7 of 39 File: USPT Oct 23, 2001

US-PAT-NO: 6306652
DOCUMENT-IDENTIFIER: US 6306652 B1

TITLE: Packaging systems for human recombinant adenovirus to be used in gene therapy

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc | Image

8. Document ID: US 6300490 B1

L6: Entry 8 of 39 File: USPT Oct 9, 2001

US-PAT-NO: 6300490
DOCUMENT-IDENTIFIER: US 6300490 B1

TITLE: Molecular constructs comprising a carcinoembryonic antigen (CEA) transcriptional regulatory region

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc | Image

9. Document ID: US 6291650 B1

L6: Entry 9 of 39 File: USPT Sep 18, 2001

US-PAT-NO: 6291650
DOCUMENT-IDENTIFIER: US 6291650 B1

TITLE: Methods for producing members of specific binding pairs

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc | Image

10. Document ID: US 6265212 B1

L6: Entry 10 of 39 File: USPT Jul 24, 2001

US-PAT-NO: 6265212
DOCUMENT-IDENTIFIER: US 6265212 B1

TITLE: Packaging systems for human recombinant adenovirus to be used in gene therapy

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc | Image

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Search Results - Record(s) 31 through 39 of 39 returned.

31. Document ID: US 5866692 A

L6: Entry 31 of 39 File: USPT Feb 2, 1999

US-PAT-NO: 5866692
DOCUMENT-IDENTIFIER: US 5866692 A

TITLE: Process for producing humanized chimera antibody

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | Claims | KWIC | Draw Desc
Image

32. Document ID: US 5858657 A

L6: Entry 32 of 39 File: USPT Jan 12, 1999

US-PAT-NO: 5858657
DOCUMENT-IDENTIFIER: US 5858657 A

TITLE: Methods for producing members of specific binding pairs

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc
Image

33. Document ID: US 5807548 A

L6: Entry 33 of 39 File: USPT Sep 15, 1998

US-PAT-NO: 5807548
DOCUMENT-IDENTIFIER: US 5807548 A

TITLE: Method of treating cancer using a chimera antibody

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc
Image

34. Document ID: US 5763266 A

L6: Entry 34 of 39 File: USPT Jun 9, 1998

US-PAT-NO: 5763266
DOCUMENT-IDENTIFIER: US 5763266 A

TITLE: Methods, compositions and devices for maintaining and growing human stem and/or

hematopoietics cells

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc | Image

35. Document ID: US 5750078 A

L6: Entry 35 of 39 File: USPT May 12, 1998

US-PAT-NO: 5750078
DOCUMENT-IDENTIFIER: US 5750078 A

TITLE: Process for producing humanized chimera antibody

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc | Image

36. Document ID: US 5710015 A

L6: Entry 36 of 39 File: USPT Jan 20, 1998

US PAT-NO: 5710015
DOCUMENT-IDENTIFIER: US 5710015 A

TITLE: cDNA cloning of inositol monophosphatase

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc | Image

37. Document ID: US 5447862 A

L6: Entry 37 of 39 File: USPT Sep 5, 1995

US-PAT-NO: 5447862
DOCUMENT-IDENTIFIER: US 5447862 A

TITLE: Pectin lyase genes of aspergillus niger

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc | Image

38. Document ID: US 5304637 A

L6: Entry 38 of 39 File: USPT Apr 19, 1994

US-PAT-NO: 5304637
DOCUMENT-IDENTIFIER: US 5304637 A

TITLE: Expression and purification of human interleukin-3 and muteins thereof

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc | Image

39. Document ID: US 5149783 A

L6: Entry 39 of 39 File: USPT Sep 22, 1992

US-PAT-NO: 5149783
DOCUMENT-IDENTIFIER: US 5149783 A

TITLE: Expression of the virally coded protease P2A of HRV2

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc | Image

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Term	Documents
CASSETTE.USPT.	55238
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Search Results - Record(s) 21 through 30 of 39 returned.

21. Document ID: US 6033908 A

L6: Entry 21 of 39 File: USPT Mar 7, 2000

US-PAT-NO: 6033908
DOCUMENT-IDENTIFIER: US 6033908 A

TITLE: Packaging systems for human recombinant adenovirus to be used in gene therapy

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc | Image

22. Document ID: US 6033668 A

L6: Entry 22 of 39 File: USPT Mar 7, 2000

US-PAT-NO: 6033668
DOCUMENT-IDENTIFIER: US 6033668 A

TITLE: Chimeric protein which confers protection against parainfluenza virus and respiratory syncytial virus

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc | Image

23. Document ID: US 6017539 A

L6: Entry 23 of 39 File: USPT Jan 25, 2000

US-PAT-NO: 6017539
DOCUMENT-IDENTIFIER: US 6017539 A

TITLE: Chimeric protein which confers protection against parainfluenza virus and respiratory syncytial virus

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc | Image

24. Document ID: US 6015708 A

L6: Entry 24 of 39 File: USPT Jan 18, 2000

US-PAT-NO: 6015708
DOCUMENT-IDENTIFIER: US 6015708 A

TITLE: Gene manipulation and expression using genomic elements

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc | Image

25. Document ID: US 5998169 A

L6: Entry 25 of 39 File: USPT Dec 7, 1999

US-PAT-NO: 5998169
DOCUMENT-IDENTIFIER: US 5998169 A

TITLE: Multimeric hybrid gene encoding a chimeric protein which confers protection against parainfluenza virus and respiratory syncytial virus

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc | Image

26. Document ID: US 5985281 A

L6: Entry 26 of 39 File: USPT Nov 16, 1999

US-PAT-NO: 5985281
DOCUMENT-IDENTIFIER: US 5985281 A

TITLE: Chemical compounds

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc | Image

27. Document ID: US 5969108 A

L6: Entry 27 of 39 File: USPT Oct 19, 1999

US-PAT-NO: 5969108
DOCUMENT-IDENTIFIER: US 5969108 A

TITLE: Methods for producing members of specific binding pairs

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc | Image

28. Document ID: US 5968776 A

L6: Entry 28 of 39 File: USPT Oct 19, 1999

US-PAT-NO: 5968776
DOCUMENT-IDENTIFIER: US 5968776 A

TITLE: Multimeric hybrid gene encoding a chimeric protein which confers protection against parainfluenza virus and respiratory syncytial virus

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc | Image

29. Document ID: US 5925565 A

L6: Entry 29 of 39 File: USPT Jul 20, 1999

US-PAT-NO: 5925565
DOCUMENT-IDENTIFIER: US 5925565 A

TITLE: Internal ribosome entry site, vector containing it and therapeutic use

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc | Image

30. Document ID: US 5871907 A

L6: Entry 30 of 39 File: USPT Feb 16, 1999

US-PAT-NO: 5871907
DOCUMENT-IDENTIFIER: US 5871907 A

TITLE: Methods for producing members of specific binding pairs

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc | Image

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Search Results - Record(s) 11 through 20 of 39 returned.

11. Document ID: US 6238893 B1

L6: Entry 11 of 39 File: USPT May 29, 2001

US-PAT-NO: 6238893
DOCUMENT-IDENTIFIER: US 6238893 B1

TITLE: Method for intracellular DNA amplification

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc | Image

12. Document ID: US 6238670 B1

L6: Entry 12 of 39 File: USPT May 29, 2001

US-PAT-NO: 6238670
DOCUMENT-IDENTIFIER: US 6238670 B1

TITLE: Compositions and methods employing a ligand for CD21 or CD19 for modulating the immune response to an antigen

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc | Image

13. Document ID: US 6225447 B1

L6: Entry 13 of 39 File: USPT May 1, 2001

US-PAT-NO: 6225447
DOCUMENT-IDENTIFIER: US 6225447 B1

TITLE: Methods for producing members of specific binding pairs

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc | Image

14. Document ID: US 6225091 B1

L6: Entry 14 of 39 File: USPT May 1, 2001

US-PAT-NO: 6225091
DOCUMENT-IDENTIFIER: US 6225091 B1

TITLE: Multimeric hybrid gene encoding a chimeric protein which confers protection against parainfluenza virus and respiratory synctial virus

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWMC | Draw Desc
Image

15. Document ID: US 6172197 B1

L6: Entry 15 of 39 File: USPT Jan 9, 2001

US-PAT-NO: 6172197
DOCUMENT-IDENTIFIER: US 6172197 B1

TITLE: Methods for producing members of specific binding pairs

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWMC | Draw Desc
Image

16. Document ID: US 6171783 B1

L6: Entry 16 of 39 File: USPT Jan 9, 2001

US-PAT-NO: 6171783
DOCUMENT-IDENTIFIER: US 6171783 B1

TITLE: Infection detection method using chimeric protein

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWMC | Draw Desc
Image

17. Document ID: US 6168786 B1

L6: Entry 17 of 39 File: USPT Jan 2, 2001

US-PAT-NO: 6168786
DOCUMENT-IDENTIFIER: US 6168786 B1

TITLE: Chimeric immunogens

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWMC | Draw Desc
Image

18. Document ID: US 6130058 A

L6: Entry 18 of 39 File: USPT Oct 10, 2000

US-PAT-NO: 6130058
DOCUMENT-IDENTIFIER: US 6130058 A

TITLE: Human NMDA R2A receptor subunit and isoforms of the human NMDA-R1 receptor subunit and encoding cDNAs

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWMC | Draw Desc | Image

19. Document ID: US 6042828 A

L6: Entry 19 of 39 File: USPT Mar 28, 2000

US-PAT-NO: 6042828
DOCUMENT-IDENTIFIER: US 6042828 A

TITLE: Humanized antibodies to ganglioside GM.sub.2

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWMC | Draw Desc | Image

20. Document ID: US 6040185 A

L6: Entry 20 of 39 File: USPT Mar 21, 2000

US-PAT-NO: 6040185
DOCUMENT-IDENTIFIER: US 6040185 A

TITLE: Genetic stabilizing elements

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWMC | Draw Desc | Image

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CASSETTE.USPT.	55238
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L6: Entry 2 of 39 File: USPT Aug 27, 2002

DOCUMENT-IDENTIFIER: US 6440422 B1
TITLE: Recombinant MVA virus, and the use thereof

Priority Application Year (1):
1995

Brief Summary Text (4):
Recombinant vaccinia virus expressing the bacteriophage T7 RNA polymerase gene allowed the establishment of widely applicable expression systems for the synthesis of recombinant proteins in mammalian cells (Moss, B., et al., Nature, 348:91-92 (1990)). In all protocols, recombinant gene expression relies on the synthesis of the T7 RNA polymerase in the cytoplasm of eukaryotic cells. Most popular became a protocol for transient-expression (Fuerst, T. R., et al., Proc. Natl. Acad. Sci. USA, 83:8122-8126 (1986) and U.S. patent application Ser. No. 7,648,971)). First, a foreign gene of interest is inserted into a plasmid under the control of the T7 RNA polymerase promoter. In the following, this plasmid is introduced into the cytoplasm of cells infected with a recombinant vaccinia virus producing T7 RNA polymerase using standard transfection procedures.

Brief Summary Text (5):
This transfection protocol is simple because no new recombinant viruses need to be made and very efficient with greater than 80% of the cells expressing the gene of interest (Elroy-Stein, O. and Moss, B., Proc. Natl. Acad. Sci. USA, 87:6743-6747 (1990)). The advantage of the vaccinia virus/T7 RNA polymerase hybrid system over other transient expression systems is very likely its independence on the transport of plasmids to the cellular nucleus. In the past, the system has been extremely useful for analytical purposes in virology and cell biology (Buonocore, L. and Rose, J. K, Nature, 345:625-628, (1990); Pattnaik, A. K and Wertz, G. W., Proc. Natl. Acad. Sci. USA, 88:1379-1383 (1991); Karschin, A. et al., FEBS Lett. 278: 229-233 (1991), Ho, B. Y. et al., FEBS Lett., 301:303-306 (1992); Buchholz, C. J. et al., Virology, 204:770-776 (1994)). However, important future applications of the vaccinia virus/T7 RNA polymerase hybrid system, as e.g. to generate recombinant proteins or recombinant viral particles for novel therapeutic or prophylactic approaches in humans, might be hindered by the productive replication of the recombinant vaccinia vector.

Brief Summary Text (13):
A recombinant MVA virus containing and capable of expressing at least one foreign gene inserted at the site of a naturally occurring deletion within the MVA genome; a recombinant MVA virus as above containing and capable of expressing at least one foreign gene inserted at the site of deletion II within the MVA genome; a recombinant MVA virus as above wherein the foreign gene codes for a marker, a therapeutic gene or an antigenic determinant; a recombinant MVA virus as above wherein the foreign gene codes for an antigenic determinant from a pathogenic virus, a bacteria, or other microorganism, or from a parasite, or a tumor cell; a recombinant MVA virus as above wherein the foreign gene codes for an antigenic determinant from Plasmodium Falciparum, Mycobacteria, Herpes virus, influenza virus, hepatitis, or human immunodeficiency viruses. a recombinant MVA virus as above wherein the antigenic determinant is HIV nef or human tyrosinase; a recombinant MVA virus as above which is MVA-LAInef or MVA-hTYR; a recombinant MVA virus as above wherein the foreign gene codes for T7 RNA polymerase; a recombinant MVA virus as above which is MVA T7 pol; a recombinant MVA virus as above wherein the foreign gene is under transcriptional control of the vaccinia virus early/late promoter P7.5; recombinant MVA viruses as

above essentially free from viruses being able to replicate in human cells; the use of a recombinant MVA virus as abcve for the transcription of DNA sequences under transcriptional control of a T7 RNA polymerase promoter; a eukaryotic cell infected by a recombinant MVA virus as any above; a cell infected by a recombinant MVA virus as above wherein the foreign gene code for T7 RNA polymerase; a cell infected by a recombinant MVA virus as above wherein the foreign gene code for T7 RNA polymerase, additionally containing one or more expression vectors carrying one or more foreign genes under transcriptional control of a T7 RNA polymerase promoter; the use of cells as above for the production of the polypeptides encoded by said foreign genes comprising: a) culturing said cells under suitable conditions, and b) isolating the polypeptides encoded by said foreign genes. a cell infected by a recombinant MVA virus as above wherein the foreign gene code for T7 RNA polymerase, additionally containing expression vectors carrying viral genes, and/or a viral vector construct encoding the genome of a viral vector under transcriptional control of a T7 RNA polymerase promoter; the use of a cells as above for the production viral particles comprising: a) culturing said cells under suitable conditions, and b) isolating the viral particles; a cell infected by a recombinant MVA virus as above wherein the foreign gene code for T7 RNA polymerase, additionally containing a) an expression vector carrying a retroviral vector construct capable of infecting and directing the expression in target cells of one or more foreign genes carried by said retroviral vector construct, and b) one or more expression vectors carrying the genes encoding the polypeptides required for the genome of said retroviral vector construct to be packaged under transcriptional control of a T7 RNA polymerase promoter; the use of cells as above for the production of retroviral particles comprising a) culturing said cells under suitable conditions, and b) isolating the retroviral particles; a vaccine containing a recombinant MVA virus as above wherein the foreign gene code for an antigenic determinant in a physiologically acceptable carrier; the use of a recombinant MVA virus as above wherein the foreign gene code for an antigenic determinant preparation of a vaccine; the use of a vaccine as above for the immunization of a living animal body, including a human; the use of a vaccine as above containing MVA-LAInef for the prevention or treatment of HIV infection or AIDS; the use of a vaccine as above containing MVA-hTYR for the prevention or treatment of melanomas; a vaccine comprising as a first component, a recombinant MVA virus as above wherein the foreign gene code for T7 RNA polymerase in a physiologically acceptable carrier, and as a second component a DNA sequence carrying an antigenic determinant under transcriptional control of a T7 RNA polymerase promoter in a physiologically acceptable carrier, the two components being contained together or separate; the use of a vaccine as above for the immunization of a living animal body, including a human, comprising inoculation of said living animal body, including a human, with the first and second component of the vaccine either simultaneously or with a timelag using the same inoculation site; and

Drawing Description Text (3):
FIG. 2 is a schematic map of pUC II LZ P7.5: MVA vector plasmid for insertion into deletion II containing P11-LacZ expression cassette and the vaccinia virus early/late promoter P7.5 to express genes of interest that can be cloned into the SmaI site of the plasmid.

Drawing Description Text (4):
FIG. 3 is a schematic map of pUCII LZdel P7.5: MVA vector plasmid for insertion of foreign genes at the site of deletion II in the MVA genome, containing a self-deleting P11-LacZ expression cassette and the vaccinia virus early/late promoter P7.5 to express genes of interest that can be cloned into the SmaI/NotI cloning site of the plasmid.

Detailed Description Text (4):
Still another object of the present invention is to provide an expression system based on a recombinant MVA virus expressing T7 RNA polymerase, and methods for the production of polypeptides, e.g. antigens or therapeutic agents, or for generating viral vectors for gene therapy or vaccines, based on this expression system.

Detailed Description Text (20):
The MVA/T7 polymerase hybrid system according to the invention can thus be used as a simple, efficient and safe mammalian expression system for production of polypeptides in the absence of productive vaccinia virus replication.

Detailed Description Text (25):
The recombinant MVA virus according to the invention expressing T7 RNA polymerase can be used to produce the proteins required for packaging retroviral vectors. To do this the gag, pol and env genes of a retrovirus (e.g. the Murine Leukemia Virus (MLV)) are placed under transcriptional control of a T7 RNA polymerase promoter in one or more expression vectors (e.g. plasmids). The expression vectors are then introduced into cells infected with the recombinant MVA virus expressing T7 RNA polymerase, together with an expression vector carrying a retroviral vector construct, possibly under transcriptional control of a T7 RNA polymerase promoter.

Detailed Description Text (27):
A further use of the recombinant MVA virus expressing T7 RNA polymerase is to generate recombinant proteins, noninfectious virus particles, or infectious mutant virus particles for the production of vaccines or therapeutics (Buchholz et al., Virology, 204:770-776 (1994) and EP-B1-1356695). To do this viral genes (e.g. the gag-pol and env genes of HIV-1) are placed under transcriptional control of the T7 promoter in an expression vector (e.g. plasmid or another recombinant MVA virus). This construct is then introduced into cells infected with the recombinant MVA virus expressing T7 RNA polymerase. The recombinant viral genes are transcribed with high efficiency, recombinant proteins are made in high amounts and can be purified. Additionally, expressed recombinant viral proteins (e.g., HIV-1 env, gag) may assemble to viral pseudo-particles that budd from the cells and can be isolated from the tissue culture medium. In another embodiment, viral proteins (from e.g. HIV, SIV, Measles virus) expressed by the MVA-T7 pol system may rescue an additionally introduced mutant virus (derived from e.g. HIV, SIV, Measles virus) by overcoming a defect in attachment and infection, uncoating, nucleic acid replication, viral gene expression, assembly, budding or another step in viral multiplication to allow production and purification of the mentioned mutant virus.

Detailed Description Text (28):
MVA-T7pol can also be used together with DNA sequences carrying the gene of an antigen of interest (e.g. the gene of HIV, nef, tat, gag, pol, or env or others) for immunization. First, a coding sequence of a given antigen (e.g HIV, HCV, HPV, HSV, measles virus, influenza virus or other) are cloned under control of a T7 RNA polymerase promoter preferably in a plasmid vector and the resulting DNA construct is amplified and purified using standard laboratory procedures. Secondly, the vector DNA is inoculated simultaneously or with appropriate timelags together with MVA-T7pol. At the site of inoculation the recombinant gene of interest is expressed transiently in cells containing both the vector DNA and MVA-T7 pol and the corresponding antigen is presented to the host immune system stimulating an antigen-specific immune response. This protocol using the non-replication vaccinia with MVA-T7 pol represents a promising novel approach to nucleic acid vaccination allowing efficient transient expression of a given antigen, but avoiding the potential risk of constitutive gene expression.

Detailed Description Text (48):
The primers for the right 550-bp DNA flank were 5'-CAG CAG CTG CAG GAA TCA TCC ATT CCA CTG AAT AGC 3' (SEQ ID NO: 3); and 5'-CAG CAG GCA TGC CGA CGA ACA AGG AAC TGT AGC AGA-3' (SEQ ID NO: 4)(sites for restriction enzymes PstI and SphI are underlined). Between these flanks of MVA DNA inserted in pUC18, the Escherichia coli LacZ gene under control of the vaccinia virus late promoter P11 (prepared by restriction digest from pIII LZ, Sutter, G. and Moss, B., PNAS USA 89:10847-10851 (1992)) was inserted, using the BamHI site, to generate the MVA insertion vector pUCII LZ (FIG. 1). In the following, a 289 bp fragment containing the vaccinia virus early-late promoter P7.5 together with a SmaI site for cloning (prepared by restriction digest with EcoRI and XbaI from the plasmid vector pSC11 (Chakrabarbti et al., Mole. Cell. Biol., 5:3403-3409 (1985)) was inserted into the SmaI site of pUCII LZ to give the MVA vector pUC II LZ P7.5 [FIG. 2]. To construct a vector plasmid that allows isolation of recombinant MVA viruses via transient synthesis of the reporter enzyme .beta.-galactosidase a 330 bp DNA fragment from the 3'-end of the E. coli LacZ open reading frame was amplified by PCR (primers were 5'-CAG CAG GTC GAC CCC GAC GGC CTT ACT GCC GCC-3' (SEQ ID NO: 5) and 5'-GGG GGG CTG CAG ATG GTA GCG ACC GGC GCT CAG-3' (SEQ ID NO: 6)) and cloned into the SalL and PstI sites of pUC II LZ P7.5 to obtain the MVA vector pUC II LZdel P7.5 (FIG. 3). Using the SmaI site, this vector plasmid

can be used to insert DNA sequences encoding a foreign gene under transcriptional control of the vaccinia virus promoter P7.5 into the MVA genome. After the desired recombinant virus has been isolated by screening for expression of .beta.-galactosidase activity further propagation of the recombinant virus leads to the self-deletion of the reengineered P11-LacZ expression cassette by homologous recombination.

Detailed Description Text (50):
A 3.1 kbp DNA fragment containing the entire gene of bacteriophage T7 RNA polymerase under control of the vaccinia virus early/late promoter P7.5 was excised with EcoRI from plasmid pTF7-3 (Fuerst, T. R. et al., P.N.A.S. USA, 83:8122-8126 (1986), modified by incubation with Klenow DNA polymerase to generate blunt ends, and cloned into a unique SmaI restriction site of pUCII LZ to make the plasmid transfer vector pUCII LZ T7pol (FIG. 4). As transcriptional regulator for the expression of the T7 RNA polymerase gene the vaccinia virus early/late promoter P7.5 was chosen. Contrary to stronger vaccinia virus late promoters (e.g. P11) this promoter system allows expression of recombinant genes immediately after the infection of target calls. The plasmid PUCII LZ T7pol that directs the insertion of the foreign-genes into the site of deletion II of the MVA genome was used to generate the recombinant virus MVA T7pol.

Detailed Description Text (52):
To monitor expression of T7 RNA polymerase by recombinant MVA T7pol [.sup.35 S] methionine -labeled polypeptides from virus infected tissue culture were analyzed. Monolayers of the monkey kidney cell line CV-1 grown in 12-well plates were infected with virus at a multiplicity of 20 TCID.sub.50 per cell. At 3 to 5 hours after infection, the medium was removed, and the cultures were washed once with 1 ml of methionine free medium. To each well, 0.2 ml of methionine-free medium supplemented with 50 .mu.Ci of [.sup.35 S] methionine was added and incubated for 30 minutes at 37.degree. C. Cytoplasmic extracts of infected cells were prepared by incubating each well in 0.2 ml of 0.5% Nonidet P-40 lysis buffer for 10 min at 37.degree. C. and samples were analyzed by SDS-PAGE. The metabolic labeling of the CV-1 cells with MVA T7pol revealed the synthesis of two additional polypeptides (i) a protein of about 116,000 Da representing the E. coli .beta.-galactosidase co-expressed to allow the screening for recombinant virus and (ii) a 98,000 Da protein with the expected size of the bacteriophage T7 RNA polymerase. The large amount of .beta.-galactocidase made by MVA T7pol is remarkable. The results from the in vivo labeling experiments demonstrate a very strong expression of the P11-LacZ gene construct when inserted into the MVA genome at the site of deletion II indicating that recombinant genes in MVA vector viruses might be expressed more efficiently when inserted into this locus of the MVA genome.

Detailed Description Text (53):
The usefulness of MVA-T7pol recombinant viruses as expression system in comparison to the WR-T7pol recombinant virus vTF7-3 (Fuerst et al. 1986) was tested by the co-transfection of DNA of a plasmid vector that is derived from pTM1 (Moss, B., et al., Nature, 348:91-92 (1990)) and contains (cloned into the NcoI and BamHI sites of the pTM1 multiple cloning site) the E. coli chloramphenicol acetyltranferase (CAT) gene under the control of a T7 RNA polymerase promoter (PT.sub.7). Transfected and infected CV-1 cells were suspended in 0.2 ml of 0.25 M Tris-HCl (pH 7.5). After three freeze-thaw cycles, the lysates were cleared by centrifugation, the protein content of the supernatants was determined, and samples containing 0.5, 0.25, 0.1 .mu.g total protein were assayed for enzyme activity as described by Mackett, M., et al., J. Virol., 49:857-864 (1984). After autoradiography, labeled spots were quantitated using the Fuji imaging analysis system.

Detailed Description Text (56):
A 648 bp DNA fragment containing the entire nef gene of HIV-1 LAI was prepared by PCR from plasmid DNA (pTG1166 kindly provided by M.-P. Kieny, Transgene S.A., Strasbourg; PCR primers were 5'-CAG CAG GGA TCC ATG GGT GGC AAG TGG TCA AAA AGT AGT-3' (SEQ ID NO: 7) and 5'-CAG CAG GGA TCC ATG TCA GCA GTT CTT GAA GTA CTC CGG-3' (SEQ ID NO: 8)), digested with restriction endonuclease BamHI, modified by incubation with Klenow DNA polymerase to generate blunt ends, and cloned into the SmaI site of pUC II LZdel P7.5 to make the vector pUC II LZdel P7.5-LAInef (FIG. 5). This plasmid could be used to engineer MVA recombinant virus that expresses the nef gene of HIV-1 LAI under control

of the vaccinia virus early/late promoter P7.5.

Other Reference Publication (11):
Wyatt, et al., "Replication-deficient vaccinia virus encoding bacteriophage T7 RNA polymerase for transient gene expression in mammalian cells", Virol., 210:202-205 (1995).